FORM PTO-1390 (Rev 5-93) U S DEPARTMENT OF COMMERCE PATENT AND TRADEMARK OFFICE

TRANSMITTAL LETTER TO THE UNITED STATES DESIGNATED/ELECTED OFFICE (DO/EO/US) CONCERNING A FILING UNDER 35 U.S.C. 371

ATTORNEY'S DOCKET NUMBER: **NITROS P146US**

U.S. APPLICATION NO. (If known, see 37 CFR 1.5): **09/673871**

INTERNATIONAL APPLICATION NO.	INTERNATIONAL FILING DATE	PRIORITY DATE CLAIMED
PCT/CH99/00163	**April 22, 1999**	**April 22, 1998**

TITLE OF INVENTION

SOLUTION FOR DIAGNOSING OR TREATING TISSUE PATHOLOGIES

APPLICANT(S) FOR DO/EO/US

Alexandre MARTI, Norbert LANGE, Matthieu ZELLWEGER, Georges WAGNIERES, Hubert VAN DEN BERGH, Patrice JICHLINSKI and Pavel KUCERA

Applicant herewith submits to the United States Designated/Elected Office (DO/EO/US) the following items and other information:

1. ■ This is a FIRST submission of items concerning a filing under 35 U.S.C. 371.
2. ☐ This is a SECOND or SUBSEQUENT submission of items concerning a filing under 35 U.S.C. 371.
3. ■ This express request to begin national examination procedures (35 U.S.C. 371(f)) at any time rather than delay examination until the expiration of the applicable time limit set in 35 U.S.C. 371(b) and PCT Articles 22 and 39(1).
4. ■ A proper Demand for International Preliminary Examination was made by the 19th month from the earliest claimed priority date.
5. ■ A copy of the International Application as filed (35 U.S.C. 371(c)(2))
   a. ☐ is transmitted herewith (required only if not transmitted by the International Bureau).
   b. ■ has been transmitted by the International Bureau. (PCT/IB/308 mailed October 28, 1999)
   c. ☐ is not required, as the application was filed in the Untied States Receiving Office (RO/US)
6. ■ A translation of the International Application into English (35 U.S.C. 371(c)(2)) is attached.
7. ■ Amendments to the claims of the International Application under PCT Article 19 (35 U.S.C. 371(c)(3))
   a. ☐ are transmitted herewith (required only if not transmitted by the International Bureau).
   b. ☐ have been transmitted by the International Bureau.
   c. ☐ have not been made; however, the time limit for making such amendments has NOT expired.
   d. ■ have not been made and will not be made.
8. ☐ A translation of the amendments to the claims under PCT Article 19 (35 U.S.C. 371(c)(3)).
9. ■ An oath or declaration of the inventor(s) (35 U.S.C. 371(c)(4)).
10. ☐ A translation of the annexes to the International Preliminary Examination Report under PCT Article 36 (35 U.S.C. 371(c)(5)).

Items 11. to 16. below concern other document(s) or information included:

11. ■ An Information Disclosure Statement under 37 CFR 1.97 and 1.98 with PTO FORM 1449.
12. ☐ An assignment document for recording. A separate cover sheet in compliance with 37 CFR 3.28 and 3.31 is included.
13. ■ A FIRST preliminary amendment.
    ☐ A SECOND or SUBSEQUENT preliminary amendment.
14. ☐ A substitute specification.
15. ☐ A change of power of attorney and/or address letter.
16. ■ Other items or information:

- ■ Preliminary Examination Report
- ☐ Annexes to Pre. Ex. Rep.
- ■ International Search Report
- ☐ French Novelty Search Report
- ☐ ___ copies of citations
- ■ Form PCT/IB/308
- ■ International Publ. No. WO 99/53962 (Face page only)
- ■ Copy of Request
- ☐ 0 sheets of formal drawings
- ■ Abstract
- ■ Verified Statement Claiming Small Entity Status
- ☐ Copy of Notification of File Missing Parts
- ☐ French Language Specification

CERTIFICATION UNDER 37 CFR 1.10

I hereby certify that this Transmittal Letter and the papers indicated as being transmitted therewith is being deposited with the United States Postal Service on this date **October 20, 2000** in an envelope as "Express Mail Post Office to Addressee" Mailing Label Number EL469353992US addressed to the: Commissioner of Patents and Trademarks, Washington, D.C. 20231.

Michael J. Bujold, Esq.
(typed or printed name of person mailing paper)

(signature of person mailing paper)

17. ■ The following fees are submitted:	CALCULATIONS	PTO USE ONLY
Basic National Fee (37 CFR 1.492(a)(1)-(5)):		
Search Report has been prepared by the EPO or JPO $860.00		
International preliminary examination fee paid to USPTO (37 CFR 1.482) $690.00		
No international preliminary examination fee paid to USPTO (37 CFR 1.482) but international search fee paid to USPTO (37 CFR 1.445(a)(2)). $710.00		
Neither international preliminary examination fee (37 CFR 1.482) nor international search fee (37 CFR 1.445(a)(2)) paid to USPTO $1000.00		
International preliminary examination fee paid to USPTO (37 CFR 1.482) and all claims satisfied provisions of PCT Article 33(1)-(4) $100.00		
ENTER APPROPRIATE BASIC FEE AMOUNT =	860	
Surcharge of $130.00 for furnishing the oath or declaration later than ☐ 20 ☐ 30 months from the earliest claimed priority date (37 CFR 1.492(e)).		

Claims	Number Filed	Number Extra	Rate		
Total Claims	9-20 =	0	x $18.00	0	
Independent Claims	1-3 =	0	x $80.00	0	
Multiple dependent claim(s) (if applicable)			+ $270.00	0	
TOTAL OF ABOVE CALCULATIONS =				860	
Reduction by 1/2 for filing by small entity, if applicable. Verified Small Entity statement must also be filed. (Note 37 CFR 1.9, 1.27, 1.28).				430	
SUBTOTAL =				430	
Processing fee of $130.00 for furnishing the English translation later the ☐ 20 ☐30 months from the earliest claimed priority date (37 CFR 1.492(f)). +					
TOTAL NATIONAL FEE =					
Fee for recording the enclosed assignment (37 CFR 1.21(h)). The assignment must be accompanied by an appropriate cover sheet (37 CFR 3.28, 3.31). $40.00 per property +				0	
TOTAL FEES ENCLOSED =				430	
				Amount to be: refunded	$
				charged	$

a. ■ A check in the amount of $ 430.00 to cover the above fees is enclosed.

b. ☐ Please charge my Deposit Account No. 04-0213 in the amount of $____ to cover the above fees. A duplicate copy of this sheet is enclosed.

c. ■ The Commissioner is hereby authorized to charge any additional fees which may be required, or credit any overpayment to Deposit Account No. 04-0213. A duplicate copy of this sheet is enclosed.

NOTE: Where an appropriate time limit under 37 CFR 1.494 or 1.495 has not been met, a petition to revive (37 CFR 1.137(a) or (b)) must be filed and granted to restore the application to pending status.

SEND ALL CORRESPONDENCE TO:

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Applicant: Alexandre MARTI et al
Serial No.:
Filed:
For: SOLUTION FOR DIAGNOSING OR TREATING TISSUE PATHOLOGIES

Attorney's Docket No.: NITROSP146US

VERIFIED STATEMENT (DECLARATION) CLAIMING SMALL ENTITY STATUS (37 CFR 1.9(f) and 1.27(b)) - INDEPENDENT INVENTOR

As a below named inventor, I hereby declare that I qualify as an independent inventor as defined in 37 CFR 1.9(c) for purposes of paying reduced fees under Section 41(a) and (b) of Title 35, United States Code, to the Patent and Trademark Office with regard to the invention entitled SOLUTION FOR DIAGNOSING OR TREATING TISSUE PATHOLOGIES described in:

[x] the specification filed herewith
[ ] application Serial No. _____, filed _____

I have not assigned, granted, conveyed or licensed and am under no obligation under contract or law to assign, grant, convey or license, any rights in the invention to any person who could not be classified as an independent inventor under 37 CFR 1.9(c) if that person had made the invention, or to any concern which would not qualify as a small business concern under 37 CFR 1.9(d) or a nonprofit organization under 37 CFR 1.9(e).

Each person, concern or organization to which I have assigned, granted, conveyed, or licensed or am under an obligation under contract or law to assign, grant, convey, or license any rights in the invention is listed below:

[x] no such person, concern, or organization
[ ] persons, concerns or organizations listed below*

*NOTE: Separate verified statements are required from each named person, concern or organization having rights to the invention averring to their status as small entities. (37 CFR 1.27)

FULL NAME _____
ADDRESS _____
[ ] INDIVIDUAL [ ] SMALL BUSINESS CONCERN [ ] NONPROFIT ORGANIZATION

FULL NAME _____
ADDRESS _____
[ ] INDIVIDUAL [ ] SMALL BUSINESS CONCERN [ ] NONPROFIT ORGANIZATION

I acknowledge the duty to file, in this application or patent, notification of any change in status resulting in loss of entitlement to small entity status prior to paying, or at the time of paying, the earliest of the issue fee or any maintenance fee due after the date on which status as a small entity is no longer appropriate. (37 CFR 1.28(b))

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application, any patent issuing thereon, or any patent to which this verified statement is directed.

WAGNIERES Georges	VAN DEN BERGH Hubert	JICHLINSKI Patrice
NAME OF INVENTOR	NAME OF INVENTOR	NAME OF INVENTOR
G. Wagnières	H. van den Bergh	P. Jichlinski
Signature of Inventor	Signature of Inventor	Signature of Inventor
5th of OCTOBER 2000	5th Oct 2000	9th oct 2000
Date	Date	Date

Applicant: Alexandre MARTI et al
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Each person, concern or organization to which I have assigned, granted, conveyed, or licensed or am under an obligation under contract or law to assign, grant, convey, or license any rights in the invention is listed below:

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I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application, any patent issuing thereon, or any patent to which this verified statement is directed.

KUCERA Pavel		
NAME OF INVENTOR	NAME OF INVENTOR	NAME OF INVENTOR
P. Kucera		
Signature of Inventor	Signature of Inventor	Signature of Inventor
6-10-2000 P. Kucera		
Date	Date	Date

Applicant: Alexandre MARTI et al
Serial No.:
Filed:
For: SOLUTION FOR DIAGNOSING OR TREATING TISSUE PATHOLOGIES

Attorney's Docket No.: NITROSP146US

VERIFIED STATEMENT (DECLARATION) CLAIMING SMALL ENTITY STATUS (37 CFR 1.9(f) and 1.27(b)) - INDEPENDENT INVENTOR

As a below named inventor, I hereby declare that I qualify as an independent inventor as defined in 37 CFR 1.9(c) for purposes of paying reduced fees under Section 41(a) and (b) of Title 35, United States Code, to the Patent and Trademark Office with regard to the invention entitled SOLUTION FOR DIAGNOSING OR TREATING TISSUE PATHOLOGIES described in:

[x] the specification filed herewith
[ ] application Serial No. _____, filed _____

I have not assigned, granted, conveyed or licensed and am under no obligation under contract or law to assign, grant, convey or license, any rights in the invention to any person who could not be classified as an independent inventor under 37 CFR 1.9(c) if that person had made the invention, or to any concern which would not qualify as a small business concern under 37 CFR 1.9(d) or a nonprofit organization under 37 CFR 1.9(e).

Each person, concern or organization to which I have assigned, granted, conveyed, or licensed or am under an obligation under contract or law to assign, grant, convey, or license any rights in the invention is listed below:

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[ ] persons, concerns or organizations listed below*

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FULL NAME _____
ADDRESS _____
[ ] INDIVIDUAL [ ] SMALL BUSINESS CONCERN [ ] NONPROFIT ORGANIZATION

FULL NAME _____
ADDRESS _____
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I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application, any patent issuing thereon, or any patent to which this verified statement is directed.

MARTI Alexandre	LANGE Norbert	ZELLWEGER Matthieu
NAME OF INVENTOR	NAME OF INVENTOR	NAME OF INVENTOR
Signature of Inventor	Signature of Inventor	Signature of Inventor
9-10-2000	05-10-2000	20/10/2000
Date	Date	Date

PATENT APPLICATION

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of	:	Alexandre MARTI, Norbert LANGE, Matthieu ZELLWEGER, Georges WAGNIERES, Hubert VAN DEN BERGH, Patrice JICHLINSKI and Pavel KUCERA
Filed	:	with an effective filing date April 22, 1999
For	:	SOLUTION FOR DIAGNOSING OR TREATING TISSUE PATHOLOGIES
Docket	:	NITROS P146US

The Commissioner of Patents and Trademarks
Washington, D.C. 20231

PRELIMINARY AMENDMENT

Dear Sir:

By way of preliminary amendment, please amend the above identified application as set forth below.

In the Claims:

Please cancel original claims 1-9, as well as any Chapter II amended claims, in favor of new claims 10 -18 as follows.

10. A 5-aminolevulinic acid ester (E-ALA) solution for producing a pharmaceutical preparation used in the diagnosis and/or treatment of tissue and/or cell lesions with local irradiation using a beam emitted by a source of light energy, which is followed, in the case of diagnosis, by detecting the fluorescence emitted by substances to which 5-aminolevulinic (ALA) or E-ALA acids are precursors, particularly protoporphyrin IX (PpIX), characterized in that the concentration C of ALA (E-ALA) ester in the solution is lower than 1% and ranges from 0.01% to 0.5%.

11. The solution according to claim 10, wherein the ALA (E-ALA) ester is ALA (h-ALA) hexylester.

12. The solution according to claim 10, wherein the solution is produced by dissolving ALA ester in a solvent compatible with the human or animal organism.

13. The solution according to claim 12, wherein said solvent is selected from the following substances: sterilized filtered water, physiological NaCl solution, phosphate buffer solution, alcohol.

14. The solution according to claim 12, wherein the solution comprises a component for adjusting the PH to a physiological value ranging from 4.8 to 8.1.

15. The solution according to claim 10, wherein the solution comprises a complementary substance to prevent the transformation of the PpIX into a heme by iron complexing in the live cells.

16. The solution according to claim 15, wherein said complementary substance is an EDTA (diaminoethyl tetra acetate).

17. The solution according to claim 15, wherein said complementary substance is deferroxamine.

18. The solution according to claim 15, wherein said complementary substance is desferal.

REMARKS

Please enter the above before consideration of this application. With respect to the above newly entered claims, please note that the subject matter of the originally filed claims is editorially revised and rewritten to bring that subject matter into conformity with the United States claim format.

In the event that there are any fee deficiencies or additional fees are payable, please charge the same or credit any overpayment to our Deposit Account (Account No. 04-0213).

Respectfully submitted,

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SOLUTION FOR DIAGNOSING OR TREATING TISSUE PATHOLOGIES

Technical Realm

The present invention concerns a 5-aminolevulinic acid ester (E-ALA) for producing a pharmaceutical preparation used in the diagnosis and treatment of tissue and/or cellular pathologies by local radiation exposure using radiation emitted by a light source followed, in the case of diagnosis, by detection of fluorescence emitted by the substances for which the 5-aminolevulinic acid ester (ALA) or the E-ALA are precursors, particularly protoporphyrin IX (PpIX).

Prior Art

The use of compounds for which ALA or ALA esters (E-ALA) and particularly hexylester hydrochloric ALA (h-ALA) are precursors is well known in the diagnosis and/or treatment of lesions, particularly cancerous lesions. This principle is thoroughly discussed in patent Publication No. WO 96/28412. The solution may be administered orally or parenterally, for example, by intra-dermal, subcutaneous, intra-peritoneal or intravenous injection. It may also be administered topically, for example locally, by exposing the surface of the organ to be treated to an E-ALA or ALA solution. A pad saturated with such a solution can also be used during topical administration. The concentration of the ALA (E-ALA) ester solution mentioned in this publication ranges from 1 to 50% and preferably between 15% and 30%. However, this concentration generates essentially no PpIX in certain organs which are principally involved in this type of treatment, namely the bladder.

In the publications in the *Journal of Photochemistry and Photobiology B and Biology* , respectively, by Fin-Puches et al entitled "Primary Clinical Response and Long-Term Follow-Up of Solar Keratoses Treated with Topically Applied 5-Aminolevulinic Acid and Irradiation by Different Wave Bands of Light," and by Chang et al entitled "The Efficacy of an Iron Chelator (CP94) in Increasing Cellular Protoporphyrin IX Following Intravestical 5-Aminolaevulinic Acid Administration: An In Vivo Study," as well as the article in *Nouvelles Dermatologiques [Dermatology News]* by P. Thomas entitled "Photothérapie

Dynamique Topique" ["Dynamic Topical Phototherapy"], the product used in treatment is ALA and not an ALA ester, which vary greatly in concentration. The ALA concentrations used are actually a minimum of 45 to 60 times higher than what is required when using an ALA ester solution (E-ALA).

Administering this substance in such strong concentrations has proven toxic to human tissue in certain instances. This toxicity, present even in the absence of light source radiation, can seriously deter generation of protoporphyrin IX (PpIX). For this reason, such concentrations either cannot be used in certain cases or are not ideal for the detection and treatment of lesions.

Furthermore, the time required to activate the active principles induced by the medicated solution is extremely long if free 5-aminolevulinic acid, that is, non-esterized ALA, is used. For this reason diagnosis and treatment using free ALA can only take place in a hospital setting, since the patient must frequently be immobilized for a very long period of time, approximately 5 hours.

In a climate where the cost of medical care is generally being reduced and preference is given to home health care, office treatment or one-day hospital care, the current treatment procedures are not only burdensome and restrictive for the patient, but costly to health insurance companies and the community.

Despite the technological progress which the use of ALA or E-ALA has contributed in terms of early diagnosis and effective treatment of certain afflictions, there are some major obstacles to its widespread use.

Description of the Invention

The goal of the present invention is to overcome these obstacles through the use of a solution designed for the diagnosis and/or treatment of cancerous lesions, particularly in the field of urology, administered in concentrations that will not prejudice biosynthesis of the active compounds and which is demonstrably very effective when applied for relatively short periods of time, making it appropriate for use in one-day clinics or even doctors' offices. Specifically, this solution must foster strong PpIX accumulation over a minimum time period and very thorough PpIX distribution throughout the treated tissue.

This goal is achieved using a 5-aminolevulinic acid ester (E-ALA) such as that defined in the preamble, characterized in that the concentration C of E-ALA in the solution is less than 1% and ranges form 0.01% to 0.5% ($0.01\% \leq C \leq 0.5\%$).

It has been shown in practice that use of a very low concentration of E-ALA in the solution increases PpIX synthesis and homogenizes distribution throughout the cellular layers, while at the same time greatly reducing secondary toxicity of the solution to the treated cells. This becomes even more important because when treating a tumor with dynamic phototherapy, the rapid photobleaching reduces PpIX concentration; complete destruction of the tumor implies an elevated initial accumulation of intracellular PpIX and thorough distribution throughout the layers of the tumor.

Advantageously, the ALA (E-ALA) ester producing the best results is hexylester hydrochloride ALA (h-ALA).

The solution is preferably produced by dissolving the ALA (E-ALA) ester in a solvent compatible with human or animal organisms.

Said solvent is advantageously selected from the following substances: sterilized filtered water, physiological NaCl solution, phosphate buffer solutions, with phosphate, or alcohol.

In its preferred form, the solution comprises a component for adjusting the PH to a physiological value ranging from 4.8 to 8.1.

In an advantageous form, the solution may comprise a complementary substance to prevent the transformation of the PpIX into a heme by iron complexing in the living cells.

Said complementary substance may be an EDTA (tetra acetate diaminoethyl), deferroaxmine or desferal.

Preferred Embodiment of the Invention

The present invention will be better understood with reference to the following description of a preferred embodiment of the solution according to the invention and its variations, and by way of illustration, a particularly

advantageous application of the solution in the diagnosis and/or treatment of lesions inside a cavity in a human or animal organism, such as the bladder.

A 5-aminolevulinic acid solution (E-ALA) is prepared by dissolving said substance, which may be an amorphous powder or in crystalline form, in an appropriate solvent compatible with *in vivo* use. By way of example, this solution may consist of sterilized demineralized water, physiological NaCl solution containing approximately 9% NaCl, a phosphate buffer solution, an alcohol, or a solution containing alcohol or the like.

This solution is preferably adjusted in PH to a value termed physiological, which depends on the application and primarily on what organ is to be treated. The PH value usually ranges from 4.8 to 8.1. If there is to be a procedure involving the bladder, the PH preferably ranges from 5.3 to 7.4.

The solution can be completed by the addition of a complementary substance to prevent the PpIX into from transforming into a heme by iron complexing in the living cells. This complementary substance may be an EDTA (tetra acetate diaminoethyl), deferroxamine or desferal.

One especially interesting application is the diagnosis and treatment of cancerous lesions in the field of urology, particularly on the interior bladder walls.

According to one application, the solution may be administered topically, contacting the interior walls of the organ. The bladder is filled with about 50 ml of low concentration ALA (E-ALA) ester or ALA (h-ALA) hexylester solution, e.g., a concentration C (by weight) ranging from 0.01% and 0.5% and preferably equal to 0.2%.

Instillation may last from ½ hour to 7 hours, but preferably ranges from ½ hour to 4 hours.

Surprisingly, it has been noted that with the use of these low concentrations which differ considerably from the 15 to 30% concentrations currently used in this field, the ALA (E-ALA) ester is more effective, as measured by an increased presence of fluorescent protoporphyrin IX (PpIX) apparent at the location of the lesions on the interior bladder walls and improved protoporphyrin distribution in the cell layers. Furthermore, due to these low concentrations,

cytotoxicity is reduced, which considerably decreases the risk of undesirable secondary effects. In particular, this reduced cytotoxicity favors the generation of the light sensitive and/or fluorescent substances to which free E-ALA or ALA are the precursors. Moreover, generating maximum PpIX shortens the time elapsing between administering the solution and performing the actual intervention.

One variation in application is defined as "fractionated topical method." It may comprise the following steps:

- a first bladder instillation lasting from ½ hour to 3 hours, and preferably lasting for about 2 hours;
- rinsing the bladder;
- a second instillation lasting from ½ hour to 3 hours, and preferably lasting for about 2 hours;
- rinsing the bladder.

After a waiting period of from 0 to 4 hours, and preferably for about 2 hours, fluorescent treatment and detection of the bladder can take place.

Topical solution of the ALA (E-ALA) ester solution or the ALA (h-ALA) hexylester solution may also be replaced by systemic application. In this case, the solution is administered either orally or parenterally either alone or in combination with compounds known as transporters, such as, for example, dimethylsulfoxide, glycine or the like, to enhance absorption and/or migration of the active substance, with the occurrence of the ALA (E-ALA) ester or the ALA (h-ALA) hexylester through the tissues and/or cells.

Finally, a way to activate penetration of the ALA (E-ALA) ester or the ALA (h-ALA) hexylester into the tissue or cells may consist of forming an iontophoresis on the walls of the organ concerned.

These phases are followed by one or more phototherapy and/or fluorescent treatment phases.

During phototherapy treatment, the walls of the organ concerned (for example, the bladder) are irradiated with a light beam called the excitant light, which may or may not be monochromatic, either continuously or sequentially,

preferably situated in the spectrum domain ranging from 300 to 900 nanometers and preferably between 350 and 650 nanometers.

During phototherapy proceedings the lighting E applied to the bladder walls, which is light power per surface unit, ranges from 0.1 mW/cm^2 to $1W/cm^2$, and preferably between $5mW/cm^2$ and $500mW/cm^2$. This light induces a phototoxic reaction due to the presence of protoporphyrin IX (PpIX) in particular and/or its photo-products in the tissue. The light doses may be applied homogeneously over the entire wall of the organ, or selectively at only the locations that have been identified as having lesions.

During fluorescent diagnosis, the bladder walls are irradiated using a beam with a spectral width ranging from 300 to 700 nanometers, and preferably from 350 to 650 nanometers. For these fluorescent diagnoses, the lighting E applied to the bladder walls (light power per surface unit) ranges from $1mW/cm^2$ and $1mW/cm^2$ and preferably between $50mW/cm^2$ to $500mW/cm^2$. The excitant light induces fluorescence in the substances to which E-ALA and especially h-ALA are precursors, particularly PpIX. This fluorescence is collected by an optical system and detected visually or by a specific, linear or matric detector such as a camera.

In addition to the advantages outlined above, the use of solutions with low ALA ester concentrations provides an inexpensive product for use in either phototherapy treatment or photodetection, at low production cost and with simplified Galenic pharmaceuticals.

Claims

1. A 5-aminolevulinic acid ester (E-ALA) solution for producing a pharmaceutical preparation used in the diagnosis and/or treatment of tissue and/or cell lesions with local irradiation using a beam emitted by a source of light energy, which is followed, in the case of diagnosis, by detecting the fluorescence emitted by substances to which 5-aminolevulinic (ALA) or E-ALA acids are precursors, particularly protoporphyrin IX (PpIX), characterized in that the concentration C of ALA (E-ALA) ester in the solution is lower than 1% and ranges from 0.01% to 0.5%.

$$0.01\% \leq C \leq 0.5\%$$

2. A solution according to claim 1 characterized in that the ALA (E-ALA) ester is ALA (h-ALA) hexylester.

3. A solution according to claim 1 characterized in that it is produced by dissolving ALA ester in a solvent compatible with the human or animal organism.

4. A solution according to claim 3 characterized in that said solvent is selected from the following substances: sterilized filtered water, physiological NaCl solution, phosphate buffer solution, alcohol.

5. A solution according to claim 3 characterized in that it comprises a component for adjusting the PH to a physiological value ranging from 4.8 to 8.1.

$$4.8 < PH < 8.1$$

6. A solution according to claim 1 characterized in that it comprises a complementary substance to prevent the transformation of the PpIX into a heme by iron complexing in the live cells.

7. A solution according to claim 6 characterized in that said complementary substance is an EDTA (diaminoethyl tetra acetate).

8. A solution according to claim 6 characterized in that said complementary substance is deferroxamine.

9. A solution according to claim 6 characterized in that said complementary substance is desferal.

SOLUTION FOR DIAGNOSING OR TREATING TISSUE PATHOLOGIES

Abstract of the Disclosure

The invention concerns a 5-aminolevulinic acid ester (E-ALA) solution for producing a pharmaceutical preparation useful for diagnosing and/or treating tissue and/or cell pathologies by local radiation exposure using radiation emitted by a light source energy followed, in the case diagnosis, by detection of fluorescent protoporphyrin IX (Pp1X). The E-ALA concentration in the solution is less than 1% and ranges between 0.01% and 0.5%. The low E-ALA concentration in the solution increases Pp1X synthesis and homogenises its distribution in the cell layers while highly reducing the secondary toxicity for the treated cells.

NITROS P146US

COMBINED DECLARATION AND POWER OF ATTORNEY

(Original, Design, National Stage of PCT, Supplemental)

As a below named inventor, I hereby declare that:

TYPE OF DECLARATION

This declaration is of the following type: (check one applicable item below)

- ☐ original
- ☐ design
- ☐ supplemental
- ■ National Stage of PCT
- ☐ divisional (see added page)
- ☐ continuation (see added page)
- ☐ continuation-in-part (see added page)

INVENTORSHIP IDENTIFICATION

My/our residence, post office address and citizenship is/are as stated below next to my/our name. I/We believe that the named inventor or inventors listed below is/are the original and first inventor or inventors of the subject matter which is claimed and for which a patent is sought on the invention entitled:

TITLE OF INVENTION

SOLUTION FOR DIAGNOSING OR TREATING TISSUE PATHOLOGIES

SPECIFICATION IDENTIFICATION

The specification of which: (complete (a), (b) or (c))

(a) ☐ is attached hereto.

(b) ☐ was filed on with an effective filing date April 22, 1999 as
☐ Serial No. _____ or
☐ Express Mail No. _____ as Serial No. (not yet known) and was amended on _____(if applicable).

(c) ■ was described and claimed in PCT International Application No. PCT/CH99/00163 filed on April 22, 1999 and as amended under PCT Article 19 on _____ (if any).

(d) ☐ amended on _____

POWER OF ATTORNEY

As a named inventor, I hereby appoint the following attorney(s) and/or agent(s) to prosecute this application and transact all business in the Patent and Trademark Office connected therewith. (list name(s) and registration number(s))

3

Anthony G. M. Davis	Registration No. 27,868
Michael J. Bujold	Registration No. 32,018
Scott A. Daniels	Registration No. 42,462

☐ Attached as part of this Declaration and Power of Attorney is the authorization of the above-named attorney(s) to accept and follow instructions from my representative(s).

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ACKNOWLEDGEMENT OF REVIEW OF PAPERS AND DUTY OF CANDOR

I hereby state that I have reviewed and understand the contents of the above identified specification, including the claims, as amended by any amendment referred to above.

I acknowledge the duty to disclose to the United States Patent Office all information which is known to be material to patentability of this application as defined in § 1.56 of Title 37 of the Code of Federal Regulations.

PRIORITY CLAIM

I hereby claim foreign priority benefits under Title 35, United States Code, § 119 of any foreign application(s) for patent or inventor's certificate or of any PCT international application(s) designating at least one country other than the United States of America listed below and have also identified below any foreign application(s) for patent or inventor's certificate or any PCT international application(s) designating at least one country other than the United States of America filed by me on the same subject matter having a filing date before that of the application(s) of which priority is claimed.

EARLIEST FOREIGN APPLICATION(S), IF ANY FILED WITHIN 12 MONTHS (6 MONTHS FOR DESIGN) PRIOR TO THIS U.S. APPLICATION

COUNTRY	APPLICATION NO.	DATE OF FILING (day,month,year)	PRIORITY CLAIMED UNDER 37 USC 119
FRANCE	98 05425	22 April 1998	☒YES ☐NO
			☐YES ☐NO
			☐YES ☐NO
			☐YES ☐NO
			☐YES ☐NO

ALL FOREIGN APPLICATION(S), IF ANY FILED MORE THAN 12 MONTHS (6 MONTHS FOR DESIGN) PRIOR TO THIS U.S. APPLICATION

DECLARATION

I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

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Inventor's signature
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Full name of **ninth** joint inventor (if any)
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Date Country of Citizenship
Residence
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